

Studies on prevalence and status of acaricide
resistance in *Rhipicephalus (Boophilus) microplus*,
a common tick in U.P.

SUMMARY

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SUMMARY

Ticks are ectoparasites of various vertebrate hosts including cattle and buffaloes. These parasites not only cause death of livestock but also responsible for reduction of quality of animal products such as milk, hide etc. Cattle and Buffaloes are the most susceptible animals for tick infestation and they contribute to maximum percentage of milk & meat in all over the world. Data regarding tick infestation revealed that, a million of livestock are dying due to the ectoparasites i.e Ticks. Further if animals survive with tick infestation, it has been observed that the animals are found to be effected severely including damaged/rough skin, pierced hide etc. Apart from which low quality milk as well as reduced milk production also are reported in infected herd. This infection further leads to many secondary infections such as small wound dermatitis, erythematous nodules erosion ulcers and many more.

Ticks are potent vectors of several disease causing agents such as bacteria, viruses, and protozoa. Ticks has occupied second position after mosquitoes for spreading pathogens in human beings, domestic and wild animals. According to a recent report on tick borne disease, India is indicated as the source country of emerging tick borne diseases. In India, around 106 species of ticks have been recorded till date. Among them *Rhipicephalus (Boophilus) microplus*, previously known as *Boophilus microplus*, is the most common cattle tick that is highly prevalent in tropical & subtropical region. They show one host life cycle including cattle, horse, sheep, goat and deer and is found to be the most prevalent cattle tick in variety of agro climatic zones of India.

The study on prevalence of ticks in U.P. state is found to be inadequate and hence there is an argent need of detailed investigation. Moreover, resistance against acaricides in ticks has become quite rampant and, hence is very prominent to carryout the research. Acaricides has adverse impact on food - chain as well as production due to their chemical nature. Hence, the present research is taken up to understand the prevalence of ticks and the areas in which resistance against acaricides is highly evident, so that techniques for integrated tick management & control may be developed and popularised.

The first chapter of the thesis includes brief description on the importance of livestock in the rural economy, parasitism its origin, morphology, life cycle, and infestation of ticks in animals. The method of diagnosis by microscopy and techniques in context of tick species *Rhipicephalus (Boophilus) microplus* along with the identification of *Hyalomma anatolicum anatolicum* and *Haemaphysalis bispinosa* also has been reported in this chapter.

The second chapter includes the literature review highlighting parasite identification, prevalence, morphological description and impact of tick infestation on their hosts, factors affecting tick growth and development as well as tick parasite interaction with its hosts considering the aims and objective of the proposed thesis.

The third chapter revealed identification and prevalence of tick parasite during the study period from January 2015 to March, 2017, in five zones (North, East, West, South and Central zone) of Uttar Pradesh state of India. The study was carried out in three seasons summer (March to June), Winter (November to February) and Rainy (July to October) season in both years.

During the study period between 2015-16, overall 2540 cattle were observed and out of which 1545 cattle were found infested with ticks and during 2016-17, 2865 cattle were examined and out of which 1790 cattle were found infected with ticks. Total prevalence was found to be 60.82% in the year 2015-16 and 62.47% prevalence was recorded for the year 2016-17. A total of 4722 (2262 ticks in 2015-16 and 2460 ticks in 2016-17) field isolates were collected from various body parts of animals during the survey period. During the research period, identification of ticks were done and tick species identified were *Rhipicephalus microplus*, *Hyalomma anatolicum anatolicum*, *Haemaphysalis bispinosa*. *Rhipicephalus microplus* was the most prevalent (Prevalence=56.01%, M:F= 1.42:1) tick species. *Hyalomma anatolicum anatolicum* was the second most abundant species which was identified, (Prevalence= 26.08%, M:F= 1.83:1), followed by *Haemaphysalis bispinosa* (Prevalence=17.9%, M:F= 1.31:1). During in the study period 2016-17, *Rhipicephalus (Boophilus) microplus* was the most prevalent tick species (Prevalence= 57.64%, M:F= 1.34:1) followed by *Hyalomma anatolicum anatolicum* (Prevalence= 28.57%, M:F= 1.35:1) and *Haemaphysalis bispinosa* (Prevalence= 1.78%, M:F= 1.51:1).

A total of 2152 animal were observed (2015- 16) out of which 1080 buffaloes were found infested with field isolates and during 2016- 17, 1275 buffaloes were found infested out of 2360. Overall abundance was found to be 50.18%, in 2015- 16 and 54.02% prevalence was during 2016- 17. In the study, during 2015-16, *Hyalomma anatolicum anatolicum* (56.02%) was found to be the most prevalent tick species followed by *Rhipicephalus (Boophilus) microplus* (43.97%) and in the year 2016-17 *Hyalomma anatolicum anatolicum* (58.97%) was found to be the most prevalent tick species followed by *Rhipicephalus (Boophilus) microplus* (41.02%). The collected ticks from the animals were dominated by male tick species.

Prevalence of tick species were done with respect to age of host. For the infestation of tick species on the body of livestock host age is an important factor. During study period (2015-16 and 2016-17) data was recorded according to the age of cattle and buffaloes for tick infestation rate in animal's body. It was observed from the data in the age group I (< 2years), highest prevalence was recorded in cattle and buffaloes (78.11% in cattle and 70.68% in young calves of buffaloes) in the year 2015-16. In the year 2016-17 highest prevalence was recorded in young calves of animal's (80.78% in cattle and 72.3% in buffaloes), followed by age group II (2-8 years) (65.73% in cattle and 62.17% in buffaloes) (66.23% in cattle and 64.93% in buffaloes). Least tick infestation was evident in III age group (>8 years) (51.66% in cattle and in buffaloes 38.52%) in year 2015-16 and in 2016 -17 tick prevalence was recorded to be 46.48% in cattle and 42.95% in buffaloes. A significant variation was found among different age groups of cattle and buffaloes ($\chi^2 = 106.33$, $p \leq 0.05$), ($\chi^2 = 151.79$, $p \leq 0.05$), respectively.

Host sex also is an important factor for the tick infestation in cattle and buffaloes. In this study, during 2015-16, 1035 male and 1505 female cattle were analyzed, out of which 425 male and 1120 female cattle were infested with the ticks. In the buffaloes, 830 male and 1322 female buffaloes were examined and out of which 370 male and 710 female buffalo were infected with ticks. High tick infestation rate in female cattle and buffaloes was observed (74.41% and 53.70%, respectively). In 2016-17, 975 male and 1890 female cattle were identified, out of which 375 male and 1415 female cattle were found to be infested with ticks. 950 male and 1410 female buffaloes were examined and out of which 445 male and 830 female buffaloes were infected with ixodid ticks. High prevalence rate was recorded in female cattle and buffaloes (74.86%, 58.86%,

respectively). Data was found to be higher in females than in the males in the year 2015-16 $\chi^2= 286.35$, $P < 0.05$ and year 2016-17 $\chi^2= 263.65$, $P < 0.05$ in the cattle) (in the year 2015-16 $\chi^2= 16.99$, $P < 0.05$ and in the year 2016-17, $\chi^2= 33.3$, $P < 0.05$ in buffaloes).

Seasonal prevalence also is one of the important factors for tick infestation examination. Tick prevalence was recorded according to three seasons; rainy, summer and winter (Rainy= July to October), (Summer= March to June) and (Winter= November to February). In the year 2015-16 a total of 890, 835 and 815 cattle were examined, out of these 703, 450 and 392 cattle were found to be infected with ticks, in rainy summer and winter season, respectively. Tick prevalence observed in rainy season (78.98%) was significantly higher followed by summer season (53.89%) and winter season (48.09%). During the study period 2016-17 prevalence of ticks was recorded higher during rainy season (73.92%), followed by summer (56.97%) and winter (55.25%). In the buffaloes, during survey period 2015-16 a total of 813, 685 and 654 buffaloes were studied and out of these 560, 300 and 220 buffaloes were found to be infested with the ticks in rainy, summer and winter season, respectively. The prevalence of ticks was recorded significantly higher in rainy season (68.88%) followed by summer (43.79%) and winter (33.63%). Significant differences were evident in prevalence of ticks among cattle and buffaloes ($\chi^2=195.47$, $p \leq 0.05$, $\chi^2=196.47$, $p \leq 0.05$, respectively). In year 2016-17, tick infestation was recorded significantly higher in both the animals, cattle and buffaloes in rainy season (67.95%), followed by summer (52.86%) and winter (41.1%). ($\chi^2= 89.06$, $p < 0.05$, $\chi^2= 116.70$, $p < 0.05$, respectively).

Prevalence of tick infestation in cattle and buffaloes differs with the rearing system of the animals. There are three type of rearing system- extensive, semi intensive and intensive rearing system and two type of rearing systems (semi intensive and intensive) are very popularly used by farmers in the study area. Rearers mostly used intensive type of rearing for calves (<2 years), weak and diseased animals. In intensively reared cattle and buffaloes higher prevalence of tick infestation was recorded (75.00%, 50.59% respectively) in the year 2015-16. In animals which were reared in semi- intensive rearing system, prevalence of tick infestation was recorded as - cattle (53.26%) and in buffaloes (47.93%). However, variations in prevalence of tick infestation in cattle and buffaloes was found significant ($\chi^2=114.31$, $P \leq 0.05$, $\chi^2=1.50$, $P \leq 0.05$ respectively). In the year 2016-17, prevalence of tick infestation was observed, significantly higher

($\chi^2=109.84$, $P\leq 0.05$, $\chi^2=10.67$, $P\leq 0.05$) in cattle and in the buffalo (76.37% and 57.73%, respectively). Semi-intensively reared animals showed variation in the prevalence of tick infestation in both cattle and buffaloes (56.01% and 51% respectively).

Breed wise tick infestation was also recorded in this research study. Local breeds includes Sahiwal, Local Zebu (cattle breeds) along with pure exotic breed Holstein Friesian, Bhadavari, Mehsana, Murrah, Surti (buffalo breeds) and cross between cross and exotic breeds were found in the areas surveyed. On the basis of origin and physical characteristics cattle breeds were determined in which during the study period (2015-16) 1237, 705 and 598 cattle 1015, 650 and 487 buffaloes were examined (Local, cross and exotic respectively) out of which 531, 423 and 474 cattle, 385, 260 and 324 buffaloes were found to be infested with ticks (42.92% local cattle breed, 60% cross breed and 79.43% exotic breed cattle) and in buffaloes prevalence was recorded to be 37.93%, 40% and 40.27% in local, cross and exotic breeds, respectively. Data was found to be significantly different in both the animals, ($\chi^2= 223.85$, $p\leq 0.05$, $\chi^2=386.32$, $p\leq 0.05$) cattle and buffaloes, respectively. During the study year, 2016-17, 1565, 680 and 620 cattle, 1315, 407 and 638 buffaloes were examined (Local, cross and exotic respectively). Out of which, 719, 490 and 581 cattle, 626, 205 and 444 buffaloes were identified to be infested with ticks. In cattle 45.94%, 72% and 93.7% (local, cross and exotic breeds respectively) tick infestation was recorded. In buffaloes 47.6%, 50.36% and 69.59% (local, cross and exotic breeds respectively) infestation of ticks was observed significantly higher in both cattle and buffaloes respectively ($\chi^2= 467.13$, $p\leq 0.05$, $\chi^2=367.62$, $p\leq 0.05$).

The main aim of the present study was to identify the correlation among the ixodid ticks in animals with respect to specific attachment site. During study period (2015-16) tick load in animals varied high on flank (19.67% in cattle and 19.01% in buffalo) followed by dewlap, perineum, groin, genitalia, ear and udder. In the year 2016-17 the tick burden in cattle was recorded to be higher in flank (16.22%) followed by udder, perineum, dewlap, groin, genitalia and ear. Tick load recorded in buffaloes was high on flank (16.22%) followed by dewlap, genitalia, udder, ear, groin and perineum.

To ensure the impact of local conditions in different zones and to know the prevalence of tick burden, differences across five zones in U.P. was studied and the results were presented. During study year 2015-16, highest prevalence (71.57%) was recorded in the cattle belonging to North zone, followed by South (60.19%), East (60%) West

(56.51%) and least in central zone (56.49%). In buffaloes North zone (67.39%), highest prevalence of ticks seen in followed by East (51.31%), West (45.56%), Central (43.62%) and least in South zone (42.55%). Zone wise prevalence of ticks in the study year 2016-17 in cattle is found to be highest in the North zone (71.92%), followed by West (70.90%), central (66.12%), south (63.44%) and least in East zone (60.51%). In the buffaloes tick prevalence was recorded highest in North zone (68.08%) followed by Central (59.53%), East (52.17%), West (47.64%) and least in South zone (44.08%).

The status of acaricide resistance in the tick species *Rhipiciphalus (Boophilus) microplus* against two commercial acaricides i.e. cypermethrin and deltamethrin was discussed in chapter four. Two bioassay tests Larval Packet Test and Adult Immersion Test was conducted to assess the status of acaricide resistance in given tick species.

Result of Larval Packet test dealt with the status of acaricide resistance in the field isolates. Tick mortality data was analysed using probit analysis. LC50 and LC95 values of cypermethrin and deltamethrin were calculated. The LC50 and LC95 values of cypermethrin for the ticks collected from different zones of Uttar Pradesh were recorded to be 29.847ppm and 422.458 ppm, respectively. Where as the values of deltamethrin were 25.106 ppm and 328.031 ppm, respectively. Regression graph was also plotted for both the chemicals and regression equation and R^2 values was also obtained.

Adult Immersion test was also performed to analyze the acaricide resistance in adult ticks. Tick mortality data was analysed by probit analysis. LC50 and LC95 values of cypermethrin and deltamethrin were calculated. The LC50 and LC95 values of cypermethrin for the collected ticks were 73.242 ppm and 522.221 ppm, respectively and that of deltamethrin were 54.553 ppm and 436.056 ppm, respectively. Regression graph was also plotted for both the chemicals and regression equation R^2 values were also obtained in the given study.

Ticks, the ectoparasites causes huge economic losses in livestock and hence on in depth analysis of its distribution and dynamics was carried out in the current research to understand and chalk out the preventive measures for its control. Tick control is usually carried out by using commercial acaricides, which leads to resistance development. Hence, the study proposes use of ethno- veterinary medicines and vaccines instead of acaricides to control prevalence of tick infestation.